



Inventor: Samir K. Brahmachari
Application No. 09/707,919
Title: METHOD OF DETECTION OF
ALLELIC VARIANTS OF SCA2 GENE
Client/Matter No. 39562-175772

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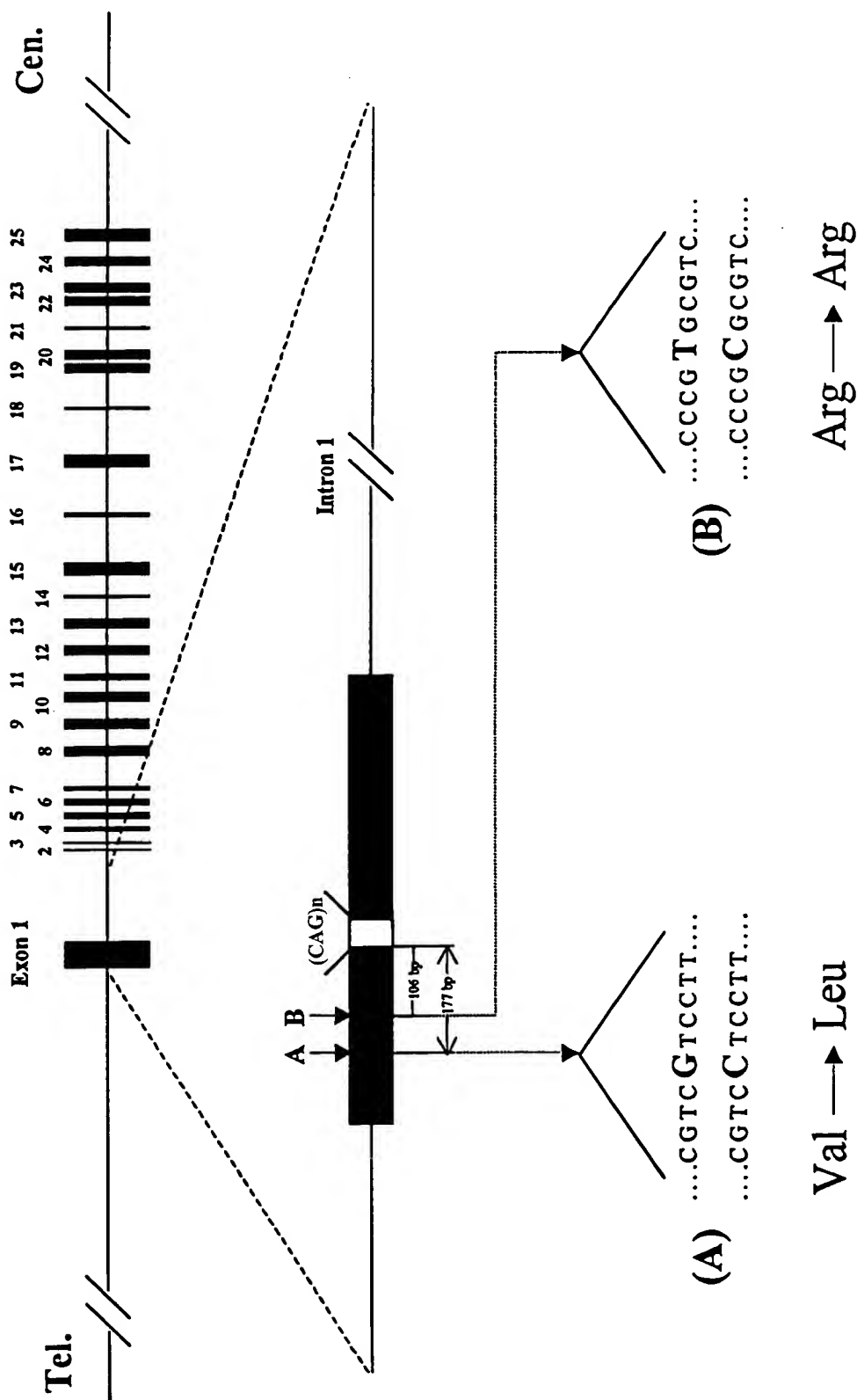


FIG 1



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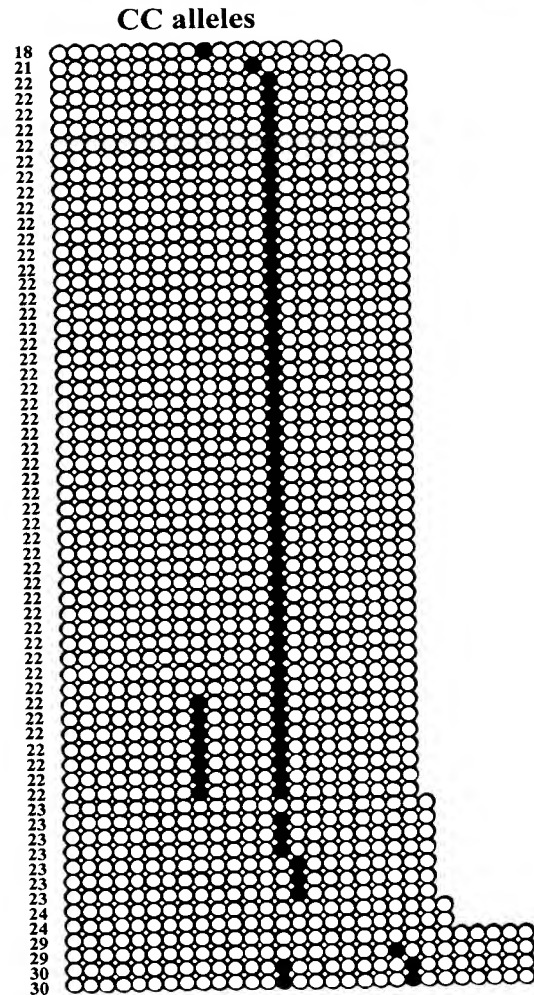
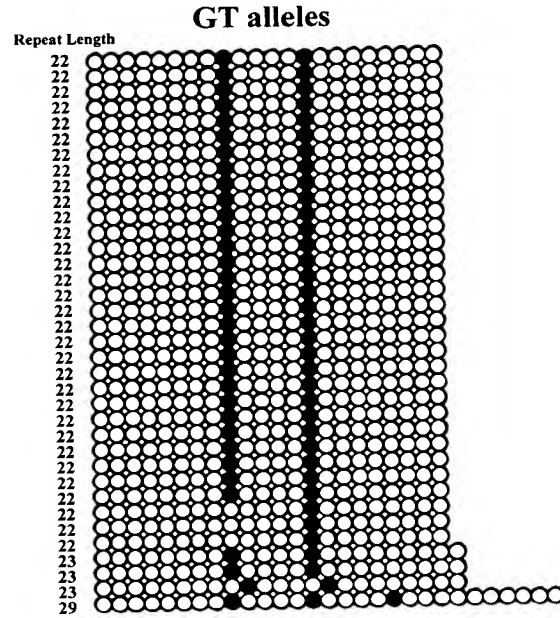
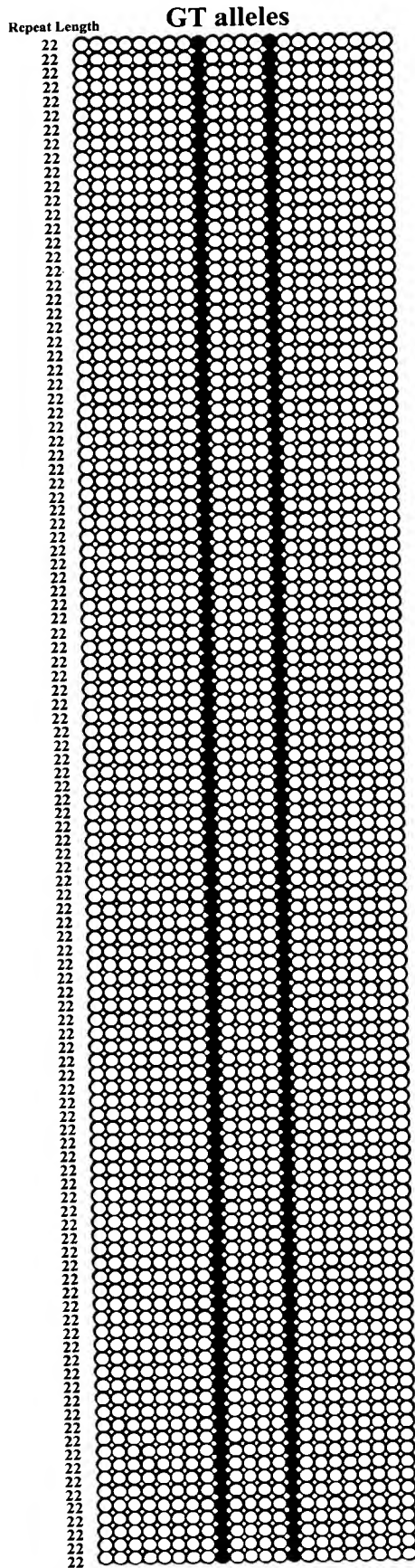


FIG 2



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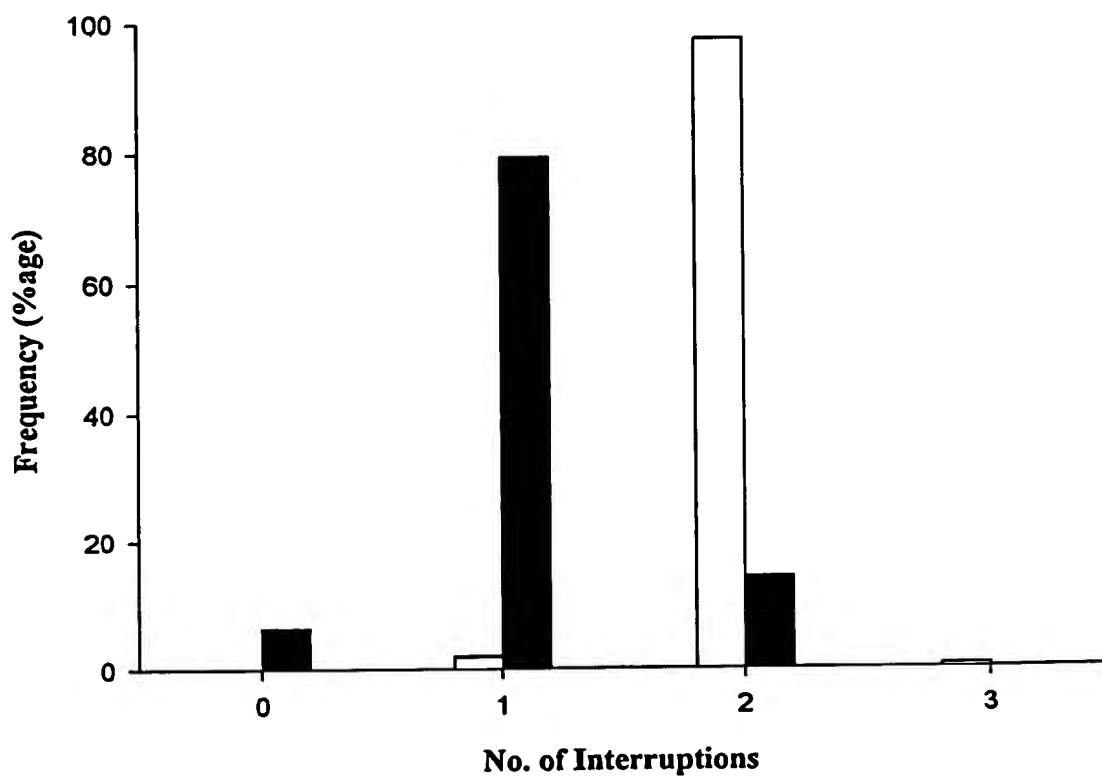


FIG. 3



Inventor: Samir K. Brahmachari
Application No. 09/707,919
Title: METHOD OF DETECTION OF
ALLELIC VARIANTS OF SCA2 GENE
Client/Matter No. 39562-175772

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SNP Details

SNP:

Handle|local_snp_id: FGU-CBT | SKB.2K.1.1
NCBI Assay Id(ss#): 869704
Reference SNP Id(rs#): 695871

STS Information: Not submitted

From SNP Database:

Submitter Handle: FGU-CBT
Submitter Batch ID: SKB.2K.1
Release Date: Aug. 2 2000 2:53PM
Molecular type: Genomic
No. of Chromosomes sampled: 215
Synonym defined:
Organism: Homo sapiens
Population: INDPOP
Submitter Method ID: SCA2-SNP
Citation:

Single Nucleotide Polymorphism in SCA2 Gene.

[View citation details](#)

NCBI Assay ID: 869704
Submitter SNP ID: SKB.2K.1.1
Synonyms:
LOCUSID: 6311
Submitter STS ID:
STS Accession: not available
GenBank Accession: U70323
Gene Name: Human ataxin-2 gene/ Spinocerebellar ataxia 2 (SCA2) gene
Length: 459

Flanking Sequence Information:

5'Assay: CTCCGCCTCA GACTGTTTTG GTAGCAACGG CAACGGCGGC GCGCGTTTC GGCCCGGCTC
CCGGCGGCTC CTTGGTCTCG GCGGGCCTCC CCGCCCCTTC GTCGTC (SEQ ID NO: 14)

Observed: G/C

3'Assay: TCCTTCTCCC CCTCGCCAGC CCGGGCGCCC CTCCGGCCGC GCCAACC CGC GCTCCCCGC
TCGGCGCCCG TCGTCCCCG CCGCGTTCCG GCGTCTCCTT GGCGCGCGCT GCTCCCGGCT
GTCCCCGCC GCGGTGCGAG CCGGTGTATG GGCCCTCAC CATGTCGCT (SEQ ID NO: 15)

3'Flank: GAAGCCCCAG CAGCAGCAGC AGCAGCAGCA GCAACAGCAG CAGCAGCAAC AGCAGCAGCA
GCAGCAGCAG CAGCCGCCGC CCGCGGCTGC CAATGTCCGC AAGCCCGGCG GCAGCGGCCT
TCTAGCGTCG CCCGCCGCC GCGCTTCGCC GTCCTCGTCC TCGGTCTCCT CGTCCTCGGC
CAC (SEQ ID NO: 16)

Allele Frequency Information:

POP Batch Id: SKB.2K.1:
Submitter Handle: FGU-CBT
Submitter Method ID: SCA2-SNP
Citation:
Single Nucleotide Polymorphism in SCA2 Gene.

Handle|PopulationID: FGU-CBT| INDPOP
No. of Chromosomes Sampled: 215

Allele: C = 0.293 / G = 0.707

FIG 4A



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Reference SNP Record

NCBI SNP ID: rs695871

NCBI Resource Links

GenBank: [U70323](#)

LocusLink: no link established

Integrated Maps: *under construction*

Submitter records for this ID:

Assay ID	Handle Local Submitter ID	Release Date
ss869704	FGU-CBT SKB.2K.1.1	Aug 2 2000 2:53PM

Variation Summary:

Assay sample size (number of chromosomes) : 430
Population data sample size (number of chromosomes) :
Total number of populations with frequency data: 1
Total number of individuals with genotype data: 0
Average estimated heterozygosity: 0.414
Average Allele Frequency:

C:	0.293
G:	0.707

Validation Summary:

Marker displays Mendelian segregation: UNKNOWN
PCR results confirmed in multiple reactions: YES
Homozygotes detected in individual genotype data: UNKNOWN
Insufficient genotype data to compute the goodness of fit to
Hardy-Weinberg
Insufficient data to compute individual x genotype consistency
measures
Validation status: *under construction*

FIG 4B



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Submitter Contact Details

This batch's contact information:

handle: FGU-CBT
name: Shweta Choudhry
fax: +91-11-7257471
phone: +91-11-7416489
email: shwetachoudhry@hotmail.com
lab: Functional genomics Unit
institution: Centre for Biochemical Technology (CSIR)
address: Delhi University Campus, Mall Road, Delhi- 110007, India

Handle information for the lab:

handle: FGU-CBT
name: Prof. Samir K. Brahmachari
fax: +91-11-7257471
phone: +91-11-7416489
email: skb@cbt.res.in
lab: Functional genomics Unit
institution: Centre for Biochemical Technology (CSIR)
address: Delhi University Campus, Mall Road, Delhi- 110007, India

FIG. 4C

SNP Population Details - 558

Submitter Population Handle: FGU-CBT
Submitter population ID: INDPOP
Population Text:

Continent: Asia
Nation: India

[View SNP used on this population](#)

FIG 4D



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SNP Method Details - 564

Submitter Method Handle: FGU-CBT
Submitter Method ID: SCA2-SNP
Method Text:

The region containing the SNP was PCR amplified using the primers SCA2-FP3 (5' CTCCGCTCAGACTGTTTGGTAG 3') (SEQ ID NO: 1) and SCA2-RP3 (5' GTGGCCGAGGACGAGGAGAC 3') (SEQ ID NO. 2). Approximately 100ng of genomic DNA was amplified in a 50ml reaction volume containing a final concentration of 5mM Tris, 25mM KCl, 0.7mM MgCl₂, 0.05% gelatin, 20pmol of each primer and 0.5U of Taq DNA polymerase. Samples were denatured at 94°C for 3 min followed by 35 cycles of denaturation (94°C, 45sec), annealing (52°C, 30sec), extension (72°C, 45sec) and a final extension of 7 min at 72°C in a Perkin Elmer GeneAmp PCR System 9600. The PCR product was purified from band cut out of the agarose gel using QIAquick gel extraction kit (Qiagen) and was directly sequenced using dye terminator chemistry on an ABI Prism 377 automated DNA sequencer with the PCR primers.

[View SNP found using this method](#)

[View SNP with population data obtained with this method](#)

FIG. 4E

SNP Publication Details

Submitter Handle: FGU-CBT
pmid:
MEDUID:
TITLE:
Single Nucleotide Polymorphism in SCA2 Gene.
AUTHOR:
CHOUDHRY, S.; BRAHMACHARI, S.K.
JOURNAL: •
VOLUME: •
SUPPL: •
ISSUE: •
I_SUPPL: •
PAGES: •
YEAR: 2000
STATUS: 1- unpulished

Searched PubMed by Author:
[CHOUDHRY, S.: BRAHMACHARI, S.K.](#)
[View SNP linked to this publication](#)

FIG. 4F



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SNP Details

SNP:

Handle|local_snp_id: FGU-CBT | SKB.2K.1.2
NCBI Assay Id(ss #): 869705
Reference SNP Id(rs #): 696872

STS Information: Not submitted

From SNP Database:

Submitter Handle: FGU-CBT
Submitter Batch ID: SKB.2K.1
Release Data: Aug 2 2000 2:53PM
Molecular type: Genomic
No. of Chromosomes sampled: 215
Synonym defined:
Organism: Homo sapiens
Population: INDPOP
Submitter Method ID: SCA2-SNP
Citation:

Single Nucleotide Polymorphism in SCA2 Gene.

[View citation details](#)

NCBI Assay ID: 869705
Submitter SNP ID: SKB.2K.1.2
Synonyms:
LOCUSID: 6311
Submitter STS ID:
STS Accession: not available
GenBank Accession: U70323
Gene Name: Human ataxin-2 gene/Sphino cerebellar ataxia 2 (SCA2) gene
Length: 459

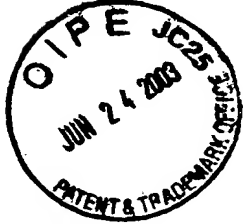
Flanking Sequence Information:

5' Assay: CTCCGCCTCA GACTGTTTTG GTAGCAACGG CAACGGCGGC GCGCGGTTTC GGCCCGGCTC
CCGGCGGCTC CTTGGTCTCG GCGGGCTCC CCGCCCTTC GTCGTCGTCC TTCTCCCCCT
CGCCAGCCCG GCGCGCCCTC CGGCCGCGCC AACCCGCGCC TCCCCGCTCG GCGCCCG
Observed: T/C (SEQ ID NO.: 17)
3' Assay: GCGTCCCCGC CGCGTTCCGG CGTCTCCTTG GCGCGCCCGG CTCCCGGCTG TCCCCGCCCCG
GCGTGCGAGC CGGTGTATGG GCCCTCACC ATGTCGCT (SEQ ID NO.: 18)
3' Assay: GAAGCCCCAG CAGCAGCAGC AGCAGCAGCA GCAACAGCAG CAGCAGCAAC AGCAGCAGCA
GCAGCAGCAG CAGCCGCCGC CCGCGGCTGC CAATGTCCGC AAGCCCGGCG GCAGCGGCCT
TCTAGCGTCG CCCGCCGCC CGCCTTCGCC GTCCTCGTCC TCGGTCTCCT CGTCCTCGGC
CAC (SEQ ID NO.: 16)

Allele Frequency Information:

POP Batch Id: SKB.2K.1:
Submitter Handle: FGU-CBT
Submitter Method ID: SCA2-SMP
Citation:
Single Nucleotide Polymorphism in SCA2 Gene.
Handle| Population ID: FGU-CBT | INDPOP
No. of Chromosomes Sampled: 215
Allele: C = 0.293 / T = 0.707

FIG 5A



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Reference SNP Record

NCBI SNP ID: rs695872

NCBI Resource Links

GenBank: U70323

LocusLink: no link established

Integrated Maps: *under construction*

Submitter records for this ID:

Assay ID	Handle Local Submitter ID	Release Date
<u>ss869705</u>	FGU-CBT SKB.2K.1.2	Aug 2 2000 2:53PM

Variation Summary:

Assay sample size (number of chromosomes): 430

Population data sample size (number of chromosomes):

Total number of populations with frequency data: 1

Total number of individuals with genotype data: 0

Average estimated heterozygosity: 0.414

Average Allele Frequency:

C: 0.293

T: 0.707

Validation Summary:

Marker displays Mendelian segregation: UNKNOWN

PCR results confirmed in multiple reactions: YES

Homozygotes detected in individual genotype data: UNKNOWN

Insufficient genotype data to compute the goodness of fit to Hardy-Weinberg

Insufficient data to compute individual x genotype consistency measures

Validation status: *under construction*

FIG. 5B



Inventor: Samir K. Brahmachari
Application No. 09/707,919
Title: METHOD OF DETECTION OF
ALLELIC VARIANTS OF SCA2 GENE
Client/Matter No. 39562-175772

10/13

SNP Publication Details

Submitter Handle:

pmid:

MEDUID:

TITLE:

Single Nucleotide Polymorphism in SCA2 Gene.

AUTHOR:

CHOUDHRY, S.; BRAHMACHARI, S. K.

JOURNAL:

VOLUME:

SUPPL:

ISSUE:

I_SUPPL:

PAGES:

YEAR: 2000

STATUS: 1- unpolished

Searched PubMed by author:

CHOUDHRY S.; BRAHMACHARI, S.K.

[View SNP linked to this publication](#)

FIG. 5C

SNP Population Details - 558

Submitter Population Handle: FCU-CBT

Submitter Population ID: INDPOP

Population Text:

Continent: Asia

Nation: India

[View SNP used on this population](#)

FIG 5D



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SNP Method Details - 564

Submitter Method Handle: FGU-CBT
Submitter Method ID: SCA2-SNP
Method Text:

The region containing the SNP was PCR amplified using the primers SCA2-FP3 (5' CTCCGCTCAGACTGTTTTGGTAG 3') (SEQ ID NO: 1) and SCA2-RP3 (5' GTGGCCGAGGACGAGGAGAC 3') (SEQ ID NO. 2). Approximately 100ng of genomic DNA was amplified in a 50ml reaction volume containing a final concentration of 5mM Tris, 25mM KCl, 0.7mM MgCl₂, 0.05% gelatin, 20pmol of each primer and 0.5U of Taq DNA polymerase. Samples were denatured at 94°C for 3 min followed by 35 cycles of denaturation (94°C, 45sec), annealing (52°C, 30sec), extension (72°C, 45sec) and a final extension of 7 min at 72°C in a Perkin Elmer GeneAmp PCR System 9600. The PCR product was purified from band cut out of the agarose gel using QIAquick gel extraction kit (Qiagen) and was directly sequenced using dye terminator chemistry on an ABI Prism 377 automated DNA sequencer with the PCR primers.

View SNP found using this method

View SNP with population data obtained with this method

FIG. 5E

Submitter Contact Details

This batch's contact information:

handle: FGU-CBT
name: Shweta Choudhry
fax: +91-11-7257471
phone: +91-11-7416489
email: shwetachoudhry@hotmail.com
lab: Functional genomics Unit
institution: Centre for Biochemical Technology (CSIR)
address: Delhi University Campus, Mall Road, Delhi- 110007, India

Handle information for the lab:

handle: FGU-CBT
name: Prof. Samir K. Brahmachari
fax: +91-11-7257471
phone: +91-11-7416489
email: skb@cbt.res.in
lab: Functional genomics Unit
institution: Centre for Biochemical Technology (CSIR)
address: Delhi University Campus, Mall Road, Delhi- 110007, India

FIG 5F



Inventor: Samir K. Brahmachari
Application No. 09/707,919
Title: METHOD OF DETECTION OF
ALLELIC VARIANTS OF SCA2 GENE
Client/Matter No. 39562-175772

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LOCUS HSU70323 4481 bp mRNA PRI 20-NOV-1996
DEFINITION Human ataxin-2 (SCA2) mRNA, complete cds.
ACCESSION U70323
VERSION U70323.1 GI:1679683
KEYWORDS
SOURCE human.
ORGANISM Homo sapiens
Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi;
Mammalia; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE 1 (bases 1 to 4481)
AUTHORS Pulst, S.-M., Nechiporuk, A., Nechiporuk, T., Gispert, S., Chen, X.-N.,
Lopes-Cendes, I., Pearlman, S., Starkman, S., Orozco-Diaz, G.,
Lunkes, A., DeJong, P., Rouleau, G.A., Auburger, G., Korenberg, J.R.,
Figueroa, C. and Sahba, S.
TITLE Moderate expansion of a normally biallelic trinucleotide repeat in
spinocerebellar ataxia type 2
JOURNAL Nat. Genet. 14 (3), 269-276 (1996)
MEDLINE 97051920
REFERENCE 2 (bases 1 to 4481)
AUTHORS Pulst, S.-M.
TITLE Direct Submission
JOURNAL Submitted (10-SEP-1996) Medicine, Cedars-Sinai, 8700 Beverly Blvd.,
Los Angeles, CA 90048, USA
FEATURES Location Qualifiers
source 1..4481
/organism="Homo sapiens"
/db_xref="taxon:9606"
/chromosome="12"
/map="12q24.1"
gene 163..4101
/gene="SCA2"
CDS 163..4101
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/codon_start=1
/product="ataxin-2"
/protein_id="AAV19200.1"
/db_xref="GI:1679684"
/translation="MRSAAAAPRSPAVATESRRFAAARWPGWRS LQRPARRSGRGGGG
AAPGPYP SAAPPPPGPGPPSRQSSPPSASDCFGSNGNGGGA FRPGSRLLGLGGPPR
PFVVVLLPLASPGAPPAAPTRASPLGARASPPRSGVSLARPAPGCP RPACFPVYGPLT
MSLKPPQQQQQQQQQQQQQQQQQQQQPPAAANVRKPGGSGLLASPA AAPSPSSSSV
SSSSATAPSSVVAATSGGGRPGLGRGRNSNKGLPQSTISFDGIYANMRMVHILTSVVG
SKCEVQVKNGGIYEGVFKTYS PKCDLVLDAAHEKSTESSSGPKREEIMESILFKCSDF
VVVQFKDMDSSYAKRDAFTDS AISAKVNGEHKEKDLEPWDAGELTANEELEALENDVS
NGWDPNDMFYRNEENYGVVSTYDSSLSSYTVPLERDNSEEF LKREARANQLAEIESS
AQYKARVALENDDRSEEEKYTA VQRNSSEREHGSINTRENKYIPPGQRNREVISWGSG
RQNSPRMGQPGSGSMP SRSTSHTSDFNPNSGSDQ RVVNGGVWPWSPCPSPSRPPSR Y
QSGPNSLPPRAATPTRPPSRPPSRPPSRPPSHPSAHGSPAPVSTMPKMSSEGPPRMSP
KAQRHPRNHRVSAGRGS ISSGLEFVSHNPPSEAATPPVARTSPSGGTWSSVVGVPRL
SPKTHRPRSPRQNSIGNTPSGPVLAS PQAGIIPTEAVAMP IPAASPTPASPNRAVT
PSSEAKDSRLQDQRQNSPAGNKENIKPNETSPSFSKAENKGISP VVSEHRKQIDDLKK
FKNDFRLQPSSTSESMDQLLNKNREGEKSRDLIKDKIEPSAKDSFIENSSS NCTSGSS
KPNSPSISPSILSNTEHKGREVT SQGVQTSSPACKQEKDDKEKKDAAEQVRKSTLN
PNAKEFNPRSFSQPKPSTTPTS PRPQAQPSPSMVGHQQT PVYTPQVCFAPNMMYPVP
VSPGVQPLYPIPMTPMPVNQAKTYRAVPNMPQQRQDQHHQS AMMHPASAAAGPPIAATP
PAYSTQYVAYSPQQFPNQPLVQHVPHYQS QHPHYVSPVIQGNARMMAPPTH AQPGLV
SSATQYGAHEQTHAMYACPKLPY NKETSPSFYFAISTGSLAQYAHPNATLHPHTPH
QPSATPTGQQQSQHGGSHPA PPSVQHHQHQAALHLAS PQQSAIYHAGLAPTPPSM
TPASNTQSPQNSFPAAQQT VFTIHPSHVQPAYTNPPHMAHVQAHVQSGMVP SHPTAH
APMMLMTTQPPGGPQAALASALQPIPVSTTAHF PYMTHPSVQAHHQQQL" (SEQ ID NO. 19)
BASE COUNT 1144 a 1380 c 1014 g 943 t

FIG 6A



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ORIGIN

```
1 accccccgaga aagcaaccca ggcgcgcgcgc cgctcctcac gtgtccctcc cggccccggg
61 gccacctcac gttctgttcc cgtctgaccc ctccgacttc cggtaaagag tccctatccg
121 cacctccgct cccacccggc gcctcggcgc gcccgccctc cgatgcgctc agcggccgca
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241 ggggtggcgt cgctccagcg gccggcgcg gggagcgggc gggggcgggc tggcgcgggc
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361 cagagctcgc ctccctccgc ctccagactgt ttggtagca acggcaacgg cggcgcgcg
421 tttcggcccg gctccggcg gctccttggt ctccggggc ctccccgccc ctctcgtctc
481 gtcttcttcc cctcgcagc cccggggccc cctccggccg cgccaacccg cgcctccccg
541 ctccggcgccc gtgcgtcccc gccgcgttcc ggctctctct tggcgcgccc ggtccccggc
601 tgtccccgccc cggcggtgca gccgggtgat gggccctca ccatgtcgt gaagccccag
661 cagcagcagc agcagcagca gcaacagcag cagcagcaac agcagcagca gcagcagcag
721 cagccggcgcc ccgcggtcgc caatgtcggc aagcccgcg gcagcggcct ctagcgtcg
781 cccgcgcgccc cgccttcgccc gctcctcgcc tgggtctcct cgctcctcgg cagcggtccc
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901 agtaacaaag gactgcctca gtctacgatt tcttttgatg gaatctatgc aaatatgagg
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1021 ggtatatatg aaggagtttt taaaacttac agtccgaagt gtgatttggg acttgatgcc
1081 gcacatgaga aaagtacaga atccagttcg gggccgaaac gtacagttta aagatatgga ctccagttat
1141 attttgttca aatgttcaga ctttgttggc gtacagttta aagtgaatgg cgaacacaaa
1201 gcaaaaagag atgtttttac tgactctgct atcagtgcct aagtgaatgg actgaggtc
1261 gagaaggacc tggagccctg ggatgcagg gaactcacag ccaatgagga actgaggtc
1321 ttggaaaaat acgtatctaa tggatggggc cccaatgata tgtttcgata taatgaagaa
1381 aattatgggt tagtgtctac gtatgatagc agtttatctt cgtatacagt gcccttagaa
1441 agagataaact cagaagaatt tttaaaacgg gaagcaaggg caaacagtt agcagaagaa
1501 attgagttcaa gtgcccagta caaagctcga gtggccctgg aaaaatgata taggagttag
1561 gaagaaaaaat acacagcagt tcagagaaat tccagtgaac gtgaggggca cagcataaac
1621 actaggggaaa ataaatatat tccctcctgga caaagaaata gagaagtcat atcctgggga
1681 agtgggagac agaattcacc gcgtatgggc cagcctggat cgggctccat gccatcaaga
1741 tccacttctc acacttcaga tttcaaccgg aattctgggt cagaccaaa agtagttaat
1801 ggagggtgtt cctggccatc gecttgcaca tctccttctc ctgcgccacc tctcgcttac
1861 cagtcagggt ccaactctct tccacctcgg gcagccaccc ctacacggcc cctccagagg
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2401 gattccaggc ttcaagatca gaggcgaac tctcctgcag ggaataaaga aaatattaaa
2461 cccaatgaaa catcacctag cttctcaaaa gctgaaaaca aaggtatatc accagttgtt
2521 tctgaacata gaaaacagat tgatgattta aagaaattta agaattgatt taggttacag
2581 ccaagttcta cttctgaatc tatggatcaa ctactaaaac ctactaagaa gggagaaaaa
2641 tcaagagatt tgatcaaaag caaaattgaa ccaagtgcct aggattcttt cattgaaaaa
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2761 atacttagta acacggagca caagagggga cctgaggtca ctcccaagg ggttcagact
2821 tccagccccc catgtaaaac agagaaagac gataagggaag agaagaaaga cgcagctgag
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2941 cagccaaaagc cttctactac cccaacttca cctcgccctc ctggttgggt tgcaccaaat
3001 atggtgggtc atcaacagcc aactccagtt tatactcagc ctgcaacctt tatacccaat
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3241 gcagccaccc caccagctta ctccagcga tatgttgctc agcatcctca tgtctatagt
3301 aatcagcccc ttgttcagca tgggccacat tatcagttct gcaccaccaa cacacgccc
3361 cctgtaatac agggtaatgc tagaatgatg gcaccaccaa cagcagcggc cagcagcggc
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4081 caccaccaa agcagttgta agctgcctct ggaagaaact aatttcattt attttgtttt
4141 ccttctactg cttctaccaa ctggaagcac ccaataagaa tgctaacagt tgcactacag
4201 taaaatatat atgttgattt cttgtaacat taggaacttg ggggctattc cataattcca
4261 ttggaagatac ttggaagcag tagagcatt ggcgaaactg gaagttattt
4321 tatgctgttt cagagtcctc aggtaccccc acatcagcta gcaaaagaag taacaagagt
4381 attttttaac aaccttgaa cttcatgaac aaaaaaaa a
4441 gattcttgct gctattactg ctaaaaaaa a
```

(SEQ ID NO: 20)

FIG 6B